factors were associated with better survival: NC at freezing (>5.5 × 10^7/kg) (HR = 0.25, p = 0.004) and FLU containing preparative regimen (HR = 0.16, p = 0.018). In patients receiving a higher cell dose, 2-y survival was 55% versus 16% for those receiving a lower dose (p = 0.005); it was 67% for those receiving a FLU containing regimen compared to 25% (p = 0.016) for those receiving other regimens. We conclude that results of UCBT for FA patients are acceptable and can be improved by better selection of the CB units and use of FLU in the preparative regimen.

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THE ROLE OF CD4$^+$CD25$^+$ T REGULATORY (TREG) CELLS IN ALLOGENEIC BONE MARROW TRANSPLANTATION

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A subpopulation of CD4$^+$ T cells that co-expresses CD25, the IL-2 receptor alpha chain, without evidence of prior activation, has been shown to be important in self-tolerance by suppressing autoimmune responses. These CD4$^+$CD25$^+$ Treg cells are present in rodents at a frequency of 8-12% of CD4$^+$ T cells and in humans at lower frequencies (1-4%). In vitro, Treg cells are potent inhibitors of alloresponses. Depletion of either host CD25+ cells prior to BMT or donor CD25+ cells in the donor graft inoculum present markedly accelerated graft-versus-host disease (GVHD) lethality. Whereas supplementing the donor graft with fresh CD4$^+$CD25$^+$ Treg cells delayed GVHD lethality, adding ex vivo activated and expanded Treg cells virtually abolished GVHD lethality. These data are consistent with the fact that activated Treg cells are more potent suppressors than non-activated cells. In studies in which CD4$^+$ effector T cells could be tracked in the whole animal, Treg cells were found to suppress effector T cell expansion or homing to lymphoid organs and GVHD target tissues. Tregs that co-expressed high levels of the lymph node homing receptor, L-selectin, were far more potent than L-selectin lo cells suggesting that homing of Tregs to secondary lymphoid organs is critical to preventing alloresponse initiation in vivo. In other studies, alloengraftment was markedly increased by the infusion of ex vivo activated and expanded donor Treg cells that expressed high levels of L-selectin. Such engraftment facilitation was not dependent upon the capacity of host T cells to receive TGFβ signals. We conclude that the infusion of ex vivo activated and expanded CD4$^+$CD25$^+$ is a highly potent means of inhibiting GVHD and facilitating alloengraftment. Based upon the known effects of Tregs on suppressing GVHD, augmenting alloengraftment and immune recovery and preserving graft-versus-leukemia effects, it is clear that a clinical trial of CD4$^+$25$^+$ merits consideration in the context of allogeneic BMT.

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IMMUNE RECONSTITUTION FOLLOWING CORD BLOOD CELL TRANSPLANTATION

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Immune reconstitution after hematopoietic stem cell transplantation (HSCT) is a fundamental problem that affects the clinical outcome of transplantation, regardless of the stem cell source. Delays or defects in immune reconstitution contribute to susceptibility to infections with viruses, fungi, and encapsulated bacteria, EBV-lymphoproliferative disease, and possibly to relapse. Ultimately, immune reconstitution depends on generation of new T lymphocytes in the thymus, a process that depends on interactions of thymocytes with thymic epithelial cells (TEC). Critical for TEC support of thymopoiesis are the cytokines IL-7 and kit ligand (KL). Umbilical cord blood cell transplantation (UCBCT) is increasingly employed because of theoretical and empiric advantages such as ready availability and decreased risk of graft-versus-host disease (GVHD). Some of the features of UCBC that make them attractive as a source of HSC may also contribute to difficulties in immune reconstitution. However, this is very difficult to analyze critically in clinical HSCT because of the large number of technical differences in transplant regimens, alloreactivity, and poorly understood variables such as donor and host variation in lymphopoiesis. One approach to understanding the outcome of UCBCT is based on analyzing the differences between the cellular components of UCBC vs other sources of stem cells. All stem cell sources contain mixtures of HSC, committed myeloid and lymphoid progenitors, and mature lymphocytes. Analyses of myeloid recovery have shown that HSC dose can be limiting, hence recommendations for a minimum number of phenotypic progenitor cells required for marrow recovery. Murine data from our laboratory suggests that thymic recovery may also be related to the dose of infused HSC or lymphoid progenitors. Although the number of HSC in UCBC sources may be limiting, the increased proliferative potential of cord blood HSC could also be advantageous. Besides reduced numbers of T lymphocytes in UCBC, a major difference between UCBC and other HSC sources, e.g., adult marrow or PBSC, is the nature of the mature T lymphocytes present in each product. The T lymphocytes in UCBC are predominantly naïve T lymphocytes while adults have mainly memory T lymphocytes. Naïve T lymphocytes require a greater amount of antigen to stimulate in vitro, compared to memory T lymphocytes. The decreased ability to stimulate naïve T cells may account for reduced alloreactivity in UCBC, but may also lead to decreased sensitivity to nominal antigens such as viruses. Thus, USCBC recipients may gain less protection from the adoptive transfer of mature T lymphocytes than BMT or PBSCT recipients. In addition, T lymphocytes from UCBC may have altered immune responses mediated by the exposure to immunomodulators in the placental microcirculation, e.g., G-CSF. Over the last five years, various studies have shown that the maintenance of mature T lymphocyte numbers in the periphery is mediated by homeostatic proliferation, in which dividing cells do not undergo activation or further maturation. The mechanism of homeostatic proliferation differs between cell types. Naïve T lymphocytes depend on self-antigen and IL-7 as signals for homeostatic proliferation. IL-7 therapy is an attractive strategy for promoting post-transplant thymopoiesis but may be complicated by exacerbation of GVHD. Recent murine experiments suggest that expansion of mature T lymphocytes by IL-7 may have a paradoxical inhibitory effect on thymopoiesis. Some approaches for translation of these experimental concepts into empirical studies in UCBCT will be described.

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INFECTIOUS DISEASE COMPLICATIONS AFTER UNRELATED DONOR HEMATOPOIETIC STEM CELL TRANSPLANTATION IN CHILDREN: IMPACT OF STEM CELL SOURCE

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Umbilical cord blood (UCB) is being increasingly used as an alternative source of hematopoietic stem cells (HSC) for unrelated donor (URD) transplantation. However, how infection risk after UCB transplantation (UCBT) compares with that seen after URD bone marrow (BM) transplantation (BMT) is not known. Therefore, we conducted a retrospective comparison of serious infectious complications in the first 2 years after HSC transplant in children transplanted for hematologic malignancy at the University of Minnesota. Our hypothesis was that due to HLA disparity, delayed neutrophil recovery, and the naivety of the neonatal immune system, UCBT recipients may be at a higher risk of both early and late infectious complications compared to recipients of unmanipulated BM. In this retrospective analysis, there were 136 children (<18 years of age) with a hematological malignancy who received cyclophosphamide 120 mg/kg and total body irradiation (TBI 1320-1375 cGy) followed by the transplantation of unmanipulated bone marrow (BM, n = 52), T cell depleted marrow (TCD, n = 24), or umbilical cord blood (n = 60). Overall, the cumulative incidence of one or more serious infections for the entire period was comparable between groups (BM 81%, TCD 83%, UCB 90%; p = 0.12). Analysis of infection density (episodes of serious infection/1000 patient days) within the time periods days 0-42, 43-100, and 101-